Article

Three new species of *Glossobalanus* (Hemichordata: Enteropneusta: Ptychoderidae) from western North America

CHRISTOPHER B. CAMERON & ANGÉLICA OSTIGUY

Sciences biologiques, Université de Montréal, C.P. 6128, Succ. Centre-ville, Montreal, QC, H3C 3J7, Canada.
E-mail: c.cameron@umontreal.ca

Abstract

Twenty three enteropneust species have been described from the west coast of North America, including one species from the family Ptychoderidae, *Glossobalanus berkeleyi* from the Salish Sea, Vancouver Island. Here we use morphology to describe three additional species of acorn worms in the genus *Glossobalanus*: *G. williami* from Cape Arago, Oregon; *G. hartmanae* and *G. barnharti* from La Jolla, California. Notes on the habit and localization of each species as well as a dichotomous key to the genera of the family Ptychoderidae are provided.

Key words: Enteropneusta, acorn worms, Ptychoderidae, dichotomous key

Introduction

The hemichordate class Enteropneusta includes about 96 species of acorn worms distributed among the families Harrimaniidae, Spengelidae, Ptychoderidae and Torquaratoridae. Acorn worms have elongated bodies ranging from less than a millimetre (Worsaae et al. 2012) to 2.5 meters in *Balanoglossus gigas*. The body is tripartite, with a proboscis, collar and trunk. The anterior proboscis is most commonly elongated or spherical. A narrow, stiff, dorsal peduncle joins the proboscis to the dorsal buccal cavity of the collar. The collar is a hollow cylinder around the mouth and is closed by contracting the proboscis against its anterior lip. The trunk can be divided externally into a branchiogenital region, a hepatic region and a caudal region. The branchiogenital region, just posterior to the collar, posseses two dorsolateral rows of gill pores and houses the serial gonads, which are highly developed in ptychoderids, forming dorsolateral genital ridges (*Glossobalanus*) or wings (*Ptychodera* and *Balanoglossus*), and in some species the gonads continue into the next two regions. The hepatic region is characterized by a pair of dorsolateral outpocketings of the gut, externally visible in *Schizocardium* and all ptychoderids. The caudal region is very fragile and sometimes bears wart-like papillae that may contain biomineralized ossicles (Cameron & Bishop 2012).

The highly specialized regions of the enteropneust gut are most evident in ptychoderids. Pharyngeal walls are perforated by a paired series of gill slits that open into branchial sacs, or atrial cavities, and these empty via gill pores to the exterior. Primary gill bars elongate and separate adjacent gill slits, and secondary gill bars, or tongue bars, grow dorsoventrally into the slit. Multiple collagenous bridges, or synaptaculae, join the primary and secondary gill bars. Primary gill bars are solid whereas tongue bars contain an extension of the trunk coelom, the peribranchial coelom. Two longitudinal ridges, the parabranchial ridges, separate the dorsal pharynx, with its gill slits, from the ventral digestive pharynx, giving the ptychoderid gut lumen a peanut shape in transverse section.

The family Ptychoderidae includes three genera: *Ptychodera*, *Balanoglossus* and *Glossobalanus*. Much of what we know about the ultrastructure of acorn worms comes from the extensive and detailed work of Jesús Benito and Fernando Pardos, who made *Glossobalanus minutus* (Fig. 1C), from the north of Spain their species of study (Benito 1975a,b; Pardos 1988; Pardos & Benito 1982, 1984, 1988a,b, 1989a,b, 1990, 1993; Benito & Pardos 1997). They characterized the cell types in the ciliated and densely glandular epidermis; the fibrillar layer of

basement membranes, including where it hypertrophies into the proboscis skeleton and gill skeletal rods; the cardiac vesicle, blood vasculature and coelomocytes; the coelomic ducts and pores; the musculature; and the cell types of the reproductive system.

Enteropneusts are dioecious and fertilization is typically external, and development is direct or via a planktonic tornaria larva. Some species can also reproduce by asexual fission and regeneration. Some of the earliest work on regeneration in hemichordates used *Glossobalanus minutus* (Dawydoff 1909, 1948). Jorge Alberto Petersen demonstrated the exceptional capacity for asexual reproduction in *G. crozieri,* a Brazilian form (Petersen & Ditadi 1971). He also performed some of the first extensive biochemical characterizations of the acorn worms (Petersen & Longhi 1971; Jorge & Petersen 1971). His tragic death, while working in the intertidal zone at São Sebastião, set back not only acorn-worm biology, but our general understanding of intertidal community ecology on the coast of Brazil. Little is known about the respiratory physiology of acorn worms; the most recent of the three studies was on *Glossobalanus crozieri* (Ditadi *et al*. 1997). Perhaps one of the most unusual features of *Glossobalanus*, being a burrowing animal, is its capacity for bioluminescence (Harvey 1922), the function of which has not been elucidated.

Here we report on three species of *Glossobalanus* from the west coast of North America. These are based on specimens from the collections of W.E. Ritter and T.H. Bullock in the Smithsonian National Museum of Natural History, Washington. For a historical account of this collection, see Deland *et al*. (2010). Our efforts to revise the alpha taxonomy of the Enteropneusta and expand on the known species from North America has, at least with the Harrimaniidae and *Schizocardium*, shed some light on the phylogenetic relationships within the families (Cameron 2002; Deland *et al*. 2010; Cameron & Perez 2012) and on the zoogeography and evolution of acorn worms in general (Cameron *et al*. 2010). Of the twelve described species of *Glossobalanus*, only one is reported from North America. To *Glossobalanus berkeleyi* reported from Vancouver Island (Willy 1931) we add an additional three species, all from the west coast. We conclude with a taxonomic key to the family Ptychoderidae.

Material and methods

Sectioned specimens of *G. williami*, *G. hartmanae* and *G. barnharti* are part of T.H. Bullock's enteropneust slide collection deposited at the Smithsonian National Museum of Natural History (USNM), Washington DC. Most of the material in this collection is fixed in Bouin's solution, with some of it fixed in formol-acetic-alcohol, and all the material was archived by transfer to 80% alcohol with 10% glycerin. Sections were cut either in paraffin or in low-viscosity nitrocellulose and mounted on glass slides. Heidenhain's iron alum haematoxylin, Masson's trichrome or Mallory's triple stain were used for staining the sections.

Systematics

Family Ptychoderidae Spengel, 1893

Ptychoderidae Spengel: van der Horst 1939.

Diagnosis. The occurrence of lateral septa in the trunk and the invariable absence of abdominal pores characterize this family. There is no vermiform process to the stomochord. Dorsal nerve roots in the collar are present and the skeletal cornua rarely reach beyond the anterior half of the collar. Synapticulae form bridges between the primary and secondary gill bars in the pharynx, and hepatic caeca are usually present. There are dorsolateral ciliated grooves in the abdominal part of the alimentary canal. The circular muscle fibers in the trunk are usually outside the longitudinal fibers. Development occurs through a typical tornaria larva.

Remarks. Species of all three genera included in this family, namely, *Ptychodera*, *Glossobalanus* and *Balanoglossus*, have been collected from the area under consideration.

Genus *Glossobalanus* Spengel, 1893

Glossobalanus Spengel: van der Horst 1939.

Diagnosis. Only genital ridges are present, never genital wings. These ridges usually extend into the hepatic region. Gill slits are small. Hepatic caeca are arranged in two regular rows. The 'cauliflower organ' is absent. (*Glossobalanus ruficollis* has neither genital ridges nor hepatic caeca, but it has a 'cauliflower organ'.) The central canals of the collar nerve cord may be well formed or may be reduced. The lateral septa are attached to the tips of the genital ridges (except in the case of *G. marginatus*). Only primary gonads occur, but both dorsal and ventral gonads are present and open to the exterior usually on the tip of the genital ridge. A ciliated streak is found only on the left side of the gut in the abdominal region (except in *G. hedleyi*, which has both the right and the left).

Remarks. We provide an augmented summary of the sole species already described from the west coast of North America, *Glossobalanus berkeleyi* Willey, 1931, and add to it new descriptions of three additional species from the same coast, *G. williami*, *G. hartmanae* (Fig. 1A) and *Glossobalanus barnharti* (Fig. 1B).
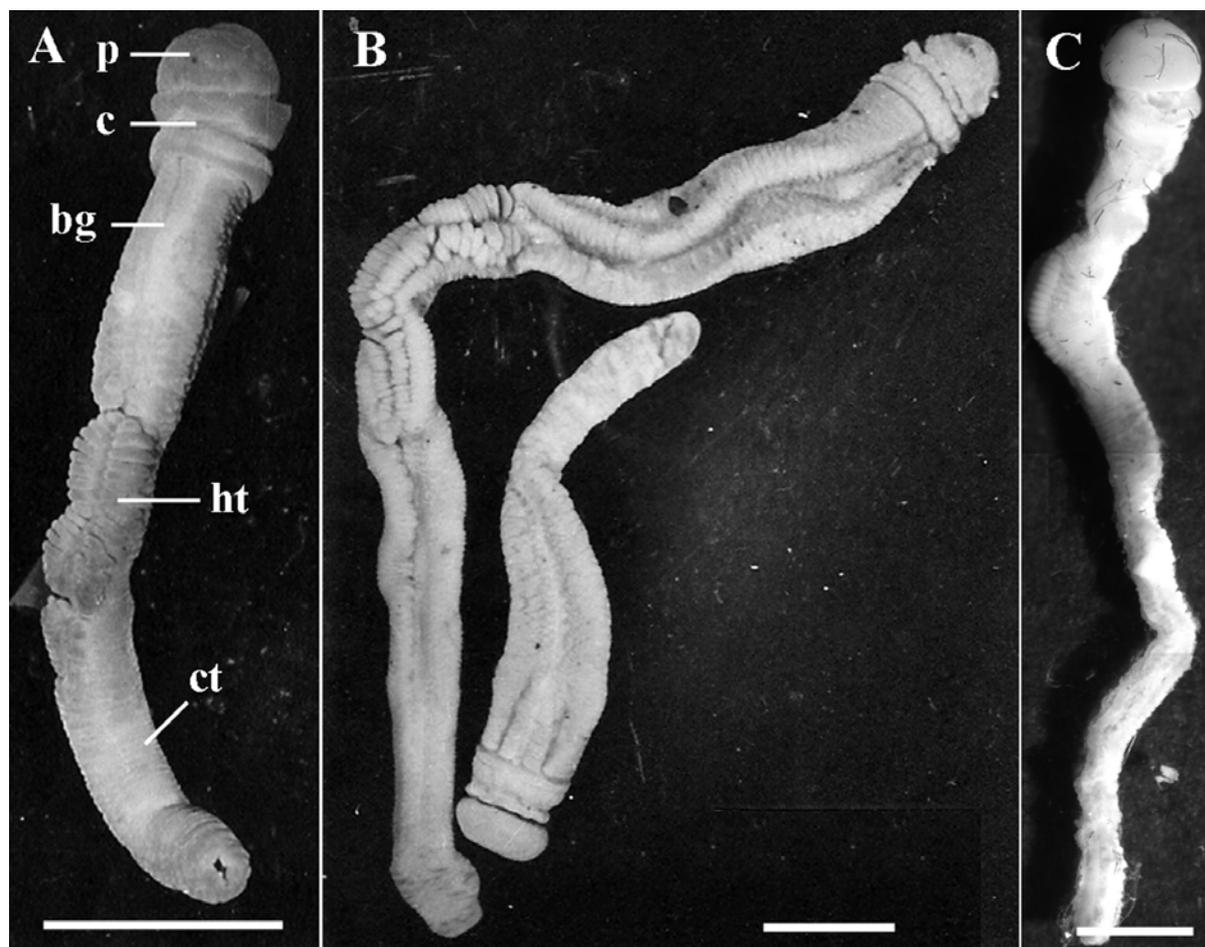

FIGURE 1. Photograph of fixed specimens of A, *Glossobalanus hartmanae* **n. sp.;** B, *G. barnharti* **n. sp.;** C, *G. minutus*. Abbeviations: **bg,** branchiogenital region of trunk; **c,** collar; **ct,** caudal region of trunk; **ht,** hepatic trunk; **p,** proboscis. Scale bar = 5 mm.

Glossobalanus berkeleyi Willey, 1931

Material examined. None; the following description is based on that of Willey (1931).

Description. Total length of only known fragment 40 mm and the dorsoventral diameter 6 mm. Proboscis, as seen in Willey's figure, rather short and only slightly longer than collar. Collar a little broader than long. Branchial region c. 18 mm long, c. 60 pairs of gill pores. Gonads (ovaries) commence anteriorly between fifth and sixth gill

pores. Genital ridges rather small (almost non-existent) in branchial region, becoming distinct beyond the pharyngeal region ("acquire the form which they possess in *G. sarniensis*" according to Willey's original description). Color in life not known.

Stomochord with a distinct lumen in the anterior part, but further posteriorly the lumen is broken up; there are two ventrolateral blind pouches for the stomochord, not directly connected with each other but independently in connection with main lumen of stomochord. Anterior part of proboscis skeleton a rounded disk-shaped structure continuous with basement membrane surrounding proboscis complex (as shown in Fig. 3 of Willey 1931). Skeletal cornua are long for a ptychoderid, extending almost to posterior end of collar. Proboscis ventral septum not extending to posterior end of ventral coelomic cavity of proboscis.

Dorsal septum of collar not complete anteriorly but begins only about middle of collar, at first dorsal root; two dorsal nerve roots in collar. Collar nerve cord with no canal, only lacunae. Anteriorly, collar nerve cord with flat dorsal surface but posteriorly almost U-shaped in cross section (as shown in Fig. 8 of Willey 1931).

Distribution. The anterior portion of only a single individual was collected near the Biological Station at Nanaimo, British Columbia, by Mrs C. Berkeley in January 1929, and was sent to Prof. Willey who described it as *G. berkeleyi* in 1931. CBC has returned to the site, which has been much modified in the past 70 years, and found no further specimens.

Glossobalanus williami n. sp.
(Fig. 2)

Etymology. The name honors G.F.G. William of Reed College, Portland, who donated several fixed specimens taken from the Oregon coast to the Bullock collection.

Material examined. *Holotype:* USNM 1192961 (T.H. Bullock accession no. 543-1), comprising nine slides. Only two specimens, one complete, were originally collected; the second is in too poor a shape to be formally designated a paratype.

Description. Whole living specimen 13 cm long, proboscis relatively short at 2.5 mm, collar 1.5 mm long. Proboscis of live specimen cream to very light tan in colour. (Data for colour and length of live worm taken from field notes made by G. Ridgel in 1958.)

Longitudinal musculature of proboscis arranged in radial bundles. Nerve-fiber layer thicker than circular-muscle-fiber layer. Ventral septum of proboscis extending nearly to tip of stomochord anteriorly but not reaching ventral wall of proboscis in this region (Fig. 2A). No dorsal septum; dorsal proboscis coelom separated into right and left only in peduncular region by cardiac vesicle. Anteriorly, cardiac vessel coextensive with stomochord. Further posteriorly, cardiac vescicle is quite roomy, reaching dorsal wall. Glomerulus very poorly developed, confined to sides, especially at junction of stomochord and cardiac vesicle (Fig. 2B). Anteriorly it extends a little beyond tip of stomochord; two halves of glomerulus confluent. Stomochord lumen extending clearly to tip. Stomochord circular in cross section in anterior region; no distinct ventral or ventrolateral blind pouches (caeca) except for moderate ventromedian bulging. Neck of stomochord with discrete lumen. Skeleton with no anterior spine, and no sharp and deep keel (Fig. 2C); a characteristic appearance in transverse section with a broad and obtuse ventral part. Only left proboscis canal present, opening out through discrete left proboscis pore (Fig. 2C). Perihaemal cavities extending a little anterior to proboscis pore. Peduncle very delicate.

Dorsal septum of collar complete anteriorly (Fig. 2D); ventral septum confined only to posterior extremity of collar. No anterior neuropore, but posterior neuropore present (Fig. 2F). Collar nerve cord lacking lumen or lacunae; no dorsal roots either. Skeletal cornua confined only to anterior third of collar, turning down sharply and extending three quarters of length down circumference of buccal cavity (Fig. 2E). Collar canals well developed, opening out along with first gill pouches (Fig. 2F).

In pharyngeal region, branchial part of pharynx occupying dorsal 2/3, with non-branchial part constituting ventral 1/3 in transverse section. Branchial openings dorsolateral, quite widely open, conspicuous and superficial (not in deep groove) (Fig. 2G). Postbranchial canal discrete, on right side of digestive part of posterior region of branchial gut. Gonads containing vitellogenic oocytes beginning only 3 mm behind collar. Only primary lateral gonads present; dorsal gonads absent. No clear-cut genital ridges, gonads lateroventral in position, appearing as single sacs (Fig. 2H). Gonadal openings ventral to gill openings.
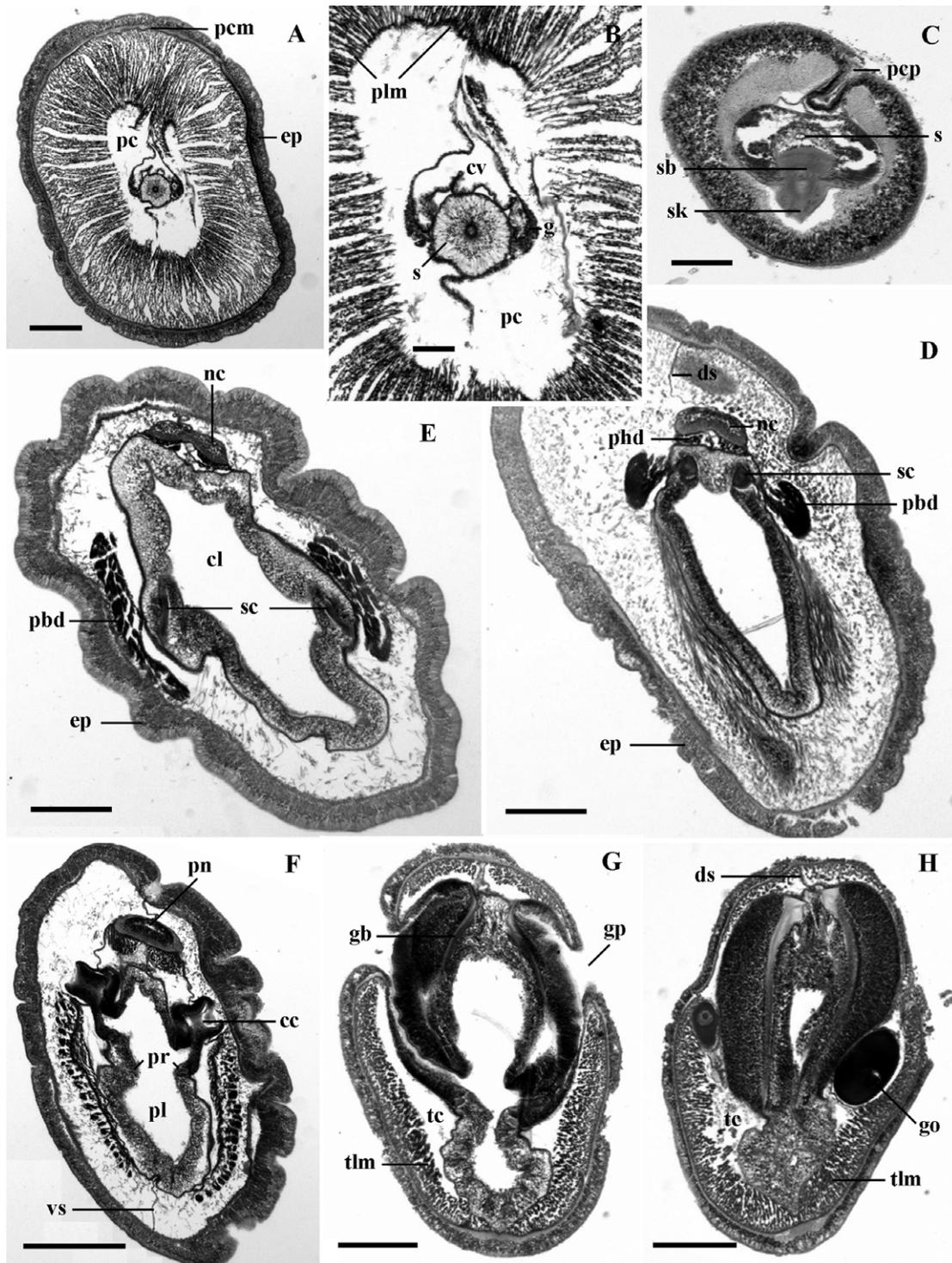
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FIGURE 2. Light micrographs of transverse sections of *Glossobalanus williami*. A, Proboscis with radial muscle bundles, heart-kidney complex, and ventral septum. B, Proboscis heart-kidney complex with a distinct lumen and poorly developed glomerulus. C, Proboscis peduncle showing the proboscis canal and obtuse skeletal keel. D, Anterior collar showing only a dorsal septum. E, Mid collar with parabuccal diverticula and lateral skeletal cornua. F, Posterior collar showing canals to the first pair of gill atria. G, Pharynx with conspicuous gill slits. H, Pharynx showing only primary gonads. Abbreviations: **cc,** collar canal; **cl**, collar lumen; **cv**, cardiac vesicle; **dep**; dorsal epidermal pocket; **ds**, dorsal septum; **ep**, epidermis **g**, glomerulus; **gb**, gill bar; **go**, gonad; **gp**, gill pore; **nc**, nerve cord; **nfl**, nerve-fiber layer; **pbd**, peribuccal diverticula; **pc**, proboscis coelom; **pcm**, proboscis circular muscles; **pcp**, proboscis canal (and pore) of the peduncle; **phd,** perihaemal diverticula; **pl**, pharynx lumen; **plm**, proboscis longitudinal muscles; **pn,** anterior neuropore; **pr,** parabranchial ridges**; s,** stomochord; **sb**, skeletal body; **sc,** skeletal cornua; **sk**, skeletal keel; **tc**, trunck coelom; **tlm**, trunk longitudinal muscles; **vs**, ventral septum. Scale bars: A, E, F = 500 μm; B, C = 100 μm; D, G, H = 250 μm.

Type locality. North Bay, Cape Arago, Coos County, Oregon. Collected by G. Ridgel 30 July 1958.

Remarks. The present form is quite distinct from the other three species occurring on the west coast of North America. It differs from both *G. hartmanae* and *G. barnharti* in the possession of the following characters: circular-fiber layer notably thicker than nerve-fiber layer in proboscis; anterior extent of glomerulus; stomochord circular in cross section anteriorly; absence of ventrolateral pouches in stomochord; almost-flat anterior face of proboscis skeleton; form of keel of skeleton; left proboscis pore; complete dorsal septum in collar; absence of dorsal nerve roots in collar; absence of lumen or lacunae in collar cord; larger size of branchial part of pharynx in transverse section; gonads starting at considerable distance posterior to collar; absence of dorsal gonads. Likewise the present form differs from *G. berkeleyi* in the following characters: absence of genital ridges; possession of discrete lumen throughout length of stomochord; absence of ventrolateral blind pouches in stomochord; very different structure and arrangement of proboscis skeleton; extent of skeletal cornua in collar; complete dorsal septum in collar; absence of dorsal nerve roots in collar; appearance of collar nerve cord in transverse section.

Glossobalanus williami is also sufficiently different from all other known *Glossobalanus* species to warrant recognition as a new species. Its defining characters are:

- very poorly developed glomerulus
- complete dorsal septum of collar
- absence of lacunae or lumen in the collar cord
- absence of dorsal roots in the collar
- no anterior neuropore, but the posterior neuropore is present
- only the primary lateral gonads present

Glossobalanus hartmanae n. sp.
(Figs 1, 3)

Etymology. The name honors Dr. Olga Hartman of the University of Southern California, long a major contributor to systematic and zoogeographic invertebrate zoology.

Material examined. *Holotype:* USNM 71750 (T.H. Bullock accession no. 206-1), comprising 12 slides.

Description. Whole animal a pale creamy white except in hepatic region, which is dark green-brown, lighter at each end. Overall length of one complete specimen 23–32 mm in medium relaxation to fair distension and 45 mm maximally extended; in the latter state, proboscis is 3.5 mm long, collar 3.5 mm, branchial region 5.5 mm, genital region 8 mm and especially distensible relative to other parts, hepatic region 4 mm in its sharply pigmented length and posthepatic abdomen about 14 mm. Genital wings not conspicuous, especially in branchial region. About 24 external hepatic saccules visible on each side externally (Fig. 1A). (Data for colour and length of live worm taken from collection cards made by T.H. Bullock in 1938.)

Epidermal epithelium of proboscis not very thick. Circular-muscle-fiber layer rather conspicuous, slightly thicker than nerve-fiber layer; longitudinal muscle fibers arranged in radial bundles, leaving a quite spacious coelom (Fig. 3A). Ventral septum extends nearly to tip of stomochord but not reaching ventral epidermis. Cardiac vesicle extending to tip of stomochord. Two halves of glomerulus rather small, confined to sides of junction between stomochord and cardiac vesicle; confluent anteriorly over tip of stomochord. Right and left dorsal proboscis canals present but narrow, opening together via single median proboscis pore. Stomochordal lumen rather narrow near anterior extremity but becoming quite marked about middle region. Stomochord with small ventrolateral pouches in communication with primary lumen (Fig. 3B). Skeleton with rather short keel anteroposteriorly, blunt, not deep dorsoventrally, stopping short of posterior end of peduncle (Fig. 3C).

Dorsal septum of collar commencing at first dorsal root, which forms anterior margin of septum; ventral septum confined to posterior fourth or fifth of collar. Midventral vascular plexus present throughout collar (Fig. 3D). Collar nerve cord only with lacunae, no central canal. Three dorsal nerve roots present (Fig. 3E). Skeletal cornua extending only half way through collar. Collar canals comprising short tubes with deep median dorsal infolding (Fig. 3E). Peribuccal cavities extending to proboscis pore, confluent at their anterior extremity. Peripharyngeal cavities extending to level where stomochord opens into buccal cavity.
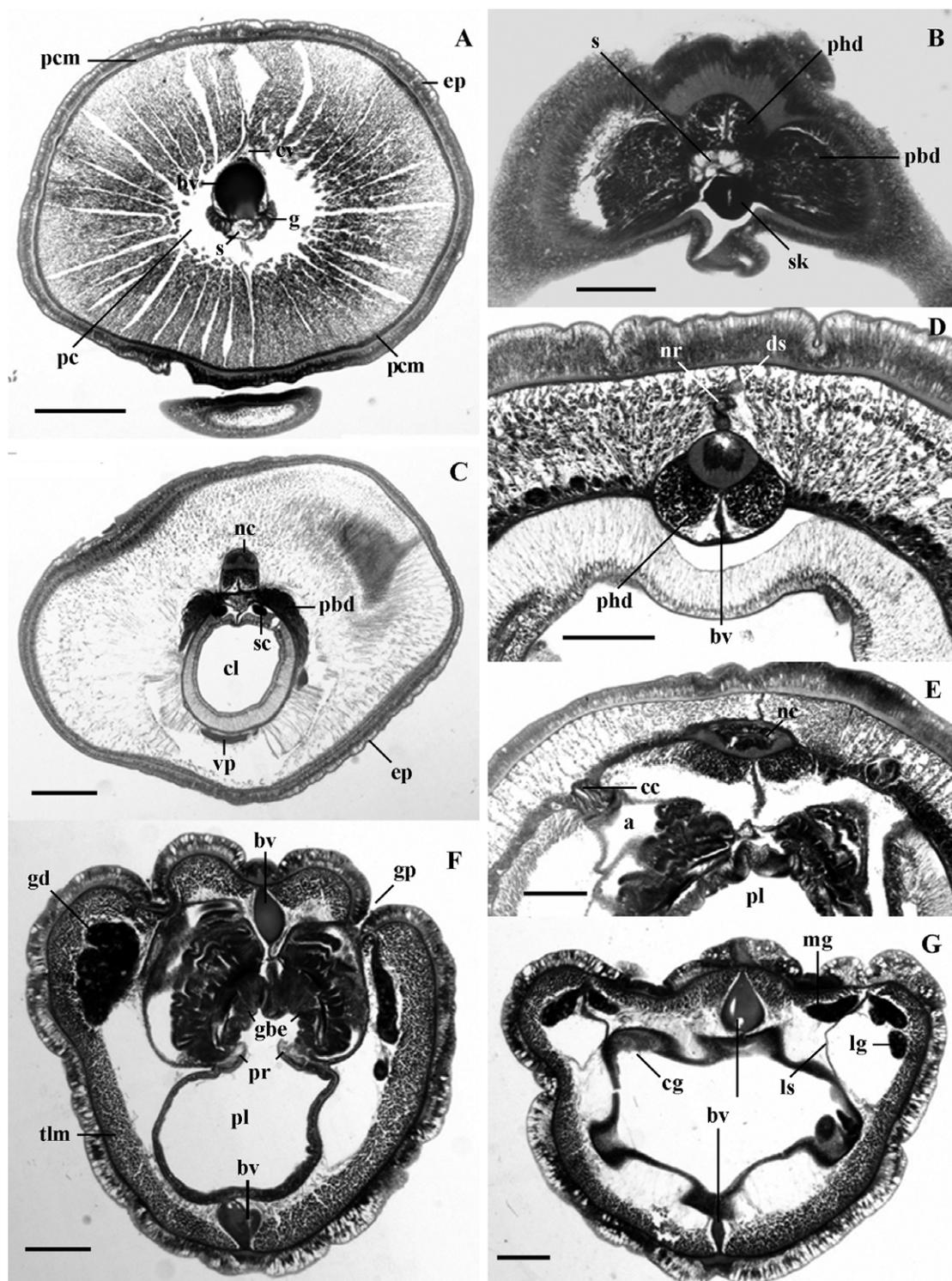
FIGURE 3. Light micrographs of transverse sections of *Glossobalanus hartmanae*. A, Proboscis with conspicuous circular-muscle layer, radial muscle bundles, heart-kidney complex and ventral septum. B, Proboscis peduncle showing the ventrolateral pouches of the stomochord and short skeletal keel. C, Collar showing a midventral vascular plexus. D, Dorsal nerve cord of collar with dorsal roots. E, Posterior collar showing canals. F, Anterior pharynx with plicate gill-bar epithelium. G, Intestinal region with lateral septum and median gonads. Abbreviations: **a,** atrium; **bv**, blood vessel; **cc,** collar canal; **cg,** ciliated groove; **cl**, collar lumen; **cv**, cardiac vesicle; **ds**, dorsal septum; **ep**, epidermis; **g**, glomerulus; **gb**, gill bar; **gbe,** gill-bar epithelia; **gd,** gonad; **gp**, gill pore; **mg**, median gonad; **lg,** lateral gonad, **ls**, lateral septum; **nc**, nerve cord; **nr**, nerve roots; **pbd,** peribuccal diverticula; **pc**, proboscis coelom; **pcm**, proboscis circular muscles; **phd,** perihaemal diverticula; **pl**, pharynx lumen; **plm**, proboscis longitudinal muscles; **pr,** parabranchial ridges; **s,** stomochord; **sc**, skeletal cornua; **sk**, skeletal keel; **tlm**, trunk longitudinal muscles; **vp**, vascular plexus. Scale bars: A, C, F, G = 500 µm; B = 200 µm; D, E = 250 µm.

Gill pouches occupying dorsal half of gut in branchial region. Outer epithelium of gill and tongue bars extremely plicate, appearing more or less like a glomerulus in section (Fig. 3F) but nuclei forming a single layer in epithelium itself. Gonads commencing immediately behind collar; only lateral primary gonads present in branchial region (Fig. 3F). Eight or nine synapticula present. Genital ridges so poorly developed as to be inconspicuous. Anteriorly, lateral septum stops short of branchial region, extending at most to penultimate gill aperture. With appearance of lateral septum median gonads also appear (Fig. 3G), these latter appearing to be more lobed than lateral gonads. Gonads extending to the commencement of hepatic region, appearing beside first one or two hepatic caeca. A short postbranchial canal present, displaced to right; abdominal ciliated groove present only on right side, commencing about middle of hepatic region. Lateral septum ceasing in anteriormost part of hepatic region.

Distribution. Five fragments including one anterior end were collected by Olga Hartman in March, 1938 at Spindrift (south end of Scripps Beach), La Jolla, California (32°51' N, 117°16' W) in crevices in shale rock. One complete specimen was collected by T.H. Bullock at the same location in December 1938.

Remarks. The description does not answer to that of any known species of *Glossobalanus*. Certain characters that are common to several other species of the genus are also present here, e.g. the anterior extent of the dorsal septum in the collar only to the first dorsal root. Like *G. ruficollis*, the present species has almost no genital ridges. Like *G. berkeleyi*, the stomochord has small ventrolateral pouches in communication with its primary lumen. But for these resemblances, the present form has no closer relationship with any known species of *Glossobalanus*. The defining characters of *Glossobalanus hartmanae* are listed below:

* right and left dorsal proboscis canals open by a single median proboscis pore
* well developed midventral vascular plexus in collar
* epithelium of the gill and tongue bars is extremely plicate
* abdominal ciliated groove is present only on the right side

Glossobalanus barnharti n. sp.
(Figs 1, 4)

Etymology. The name honors Dr. Percy S. Barnhart, long-time curator of the Scripps Institution of Oceanography.
Material examined. *Holotype:* USNM 71749 (T.H. Bullock accession no. 207-1), comprising 29 slides.
Description. Proboscis twice as long as its base width. Collar broader than long, with shallow groove ringing its middle. Genital ridges beginning immediately behind collar. Branchiogenital region as in *G. minutus* (Fig. 1C). Circular-muscle-fiber layer of proboscis c. 1.5 times thickness of the nerve-fiber layer; longitudinal muscle fibers arranged in massive radial groups (Fig. 4A). Ventral septum extending nearly to tip of stomochord. Cardiac vesicle nearly coextensive with stomochord anteriorly. Right and left halves of glomerulus confluent at anterior extremity. Dorsal glomerulus small. Stomochordal lumen narrow, well defined to tip. Forepart of stomochord flattened, watchglass-like in transverse sections (Fig. 4B). Stomochord with single midventral blind pouch (Fig. 4C). Only left dorsal proboscis coelom opens to exterior by median dorsal proboscis pore, with spacious (bulbous) medially placed proboscis canal (vesicle). Anteriorly, body and keel of skeleton parted by invasion of coelomic epithelium, forming a spacious cavity that narrows posteriorly, stopping about level with proboscis pore. From here, posteriorly in peduncle, keel is rather deep and narrow (Fig. 4D); further posteriorly it is shortened dorsoventrally, but extends some distance into collar region.

Dorsal septum of collar extending anteriorly to first dorsal root; ventral septum absent. Midventral vascular plexus of collar poorly developed (Fig. 4E). Cornua extending half way through collar, taking sharp turn ventrally, ending about midway on lateral walls of buccal cavity. Collar nerve cord rather flattened, lateral margins thicker than rest. No axial canal, only some large lacunae. Anterior neuropore present. Eight or nine dorsal roots (Fig. 4F). Perihaemal cavities extending to proboscis pore, confluent at their anterior extremity. Peribuccal cavities extending to about level of origin of skeletal cornua. Collar canals with deep dorsal fold.

Gonads commencing immediately behind collar. Dorsal pharynx larger than ventral digestive pharynx (Fig. 4G). Lateral septum extending anteriorly to some distance into posterior part of branchial region. Lateral septum and medial gonads present. Presence of post-branchial canal not determined. Gonads continuing well into intestinal region (Fig. 4H).
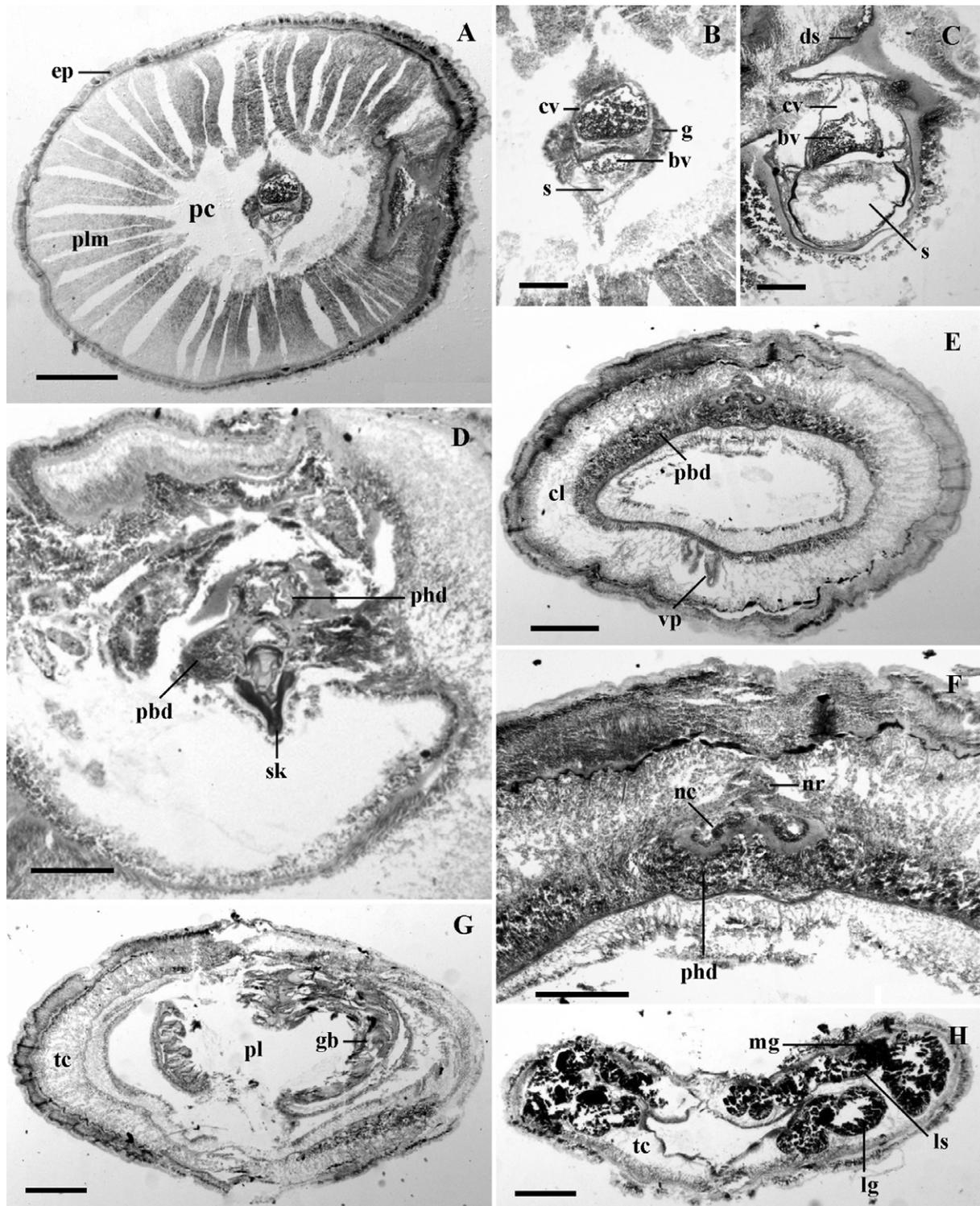
FIGURE 4. Light micrographs of transverse sections of *Glossobalanus barnharti*. A, Proboscis with massive radial muscle bundles. B, Proboscis heart-kidney complex with lateral and dorsal glomerulus. C, Peduncle showing the large lumen of the stomochord. D, Peduncle with deep and narrow keel. E, Collar. F, Collar cord with dorsal nerve roots. G, Anterior pharynx large dorsal branchial lumen. H, Intestinal region with lateral septum and median gonads. Abbreviations: **bv**, blood vessel; **cc,** collar canal; **cl**, collar lumen; **clm**, collar longitudinal muscles; **cv**, cardiac vesicle; **ds**, dorsal septum; **ep**, epidermis **g**, glomerulus; **gb**, gill bar; **mg**, median gonad; **lg,** lateral gonad; **ls**, lateral septum; **mg**, median gonad; **nc**, nerve cord; **nr**, nerve roots; **pbd,** peribuccal diverticula; **pc**, proboscis coelom; **phd,** perihaemal diverticula; **pl**, pharynx lumen; **plm**, proboscis longitudinal muscles; **s**, stomochord; **sk**, skeletal keel; **tc**, trunk coelom; **vp**, vascular plexus. Scale bars: A, E, F, G, H = 500 μm; B, C = 200 μm; (D) = 250 μm.

Distribution. Two specimens, one apparently complete, were taken by P.S. Barnhart (his acc. No. 1195) from the intertidal zone of the protected outer coast at Bird Rock, La Jolla, California (32°49' N, 117°14' W), in June 1921.

Remarks. It has already been stated above that the form under consideration resembles to some extent *G. minutus* in its external appearance. But in its internal anatomy it does not quite resemble that species. The two agree only in a few specific characters such as the forward extension of the proboscis ventral septum, the presence of a dorsal glomerulus and the absence of the ventral septum in the collar. The present species appears anatomically to be more closely related to *G. sarniensis* than to any other species of the genus. Thus these two have the following characters in common: connection of the left dorsal proboscis coelom with a large bulbous, median proboscis canal; continuous lumen in stomochord; cardiac vesicle anteriorly coextensive with stomochord; large keel and the separation of the keel and the body in the anterior part of the skeleton by a coelomic invasion. They differ in many more characters.

In external appearance *G. barnharti* differs from *G. sarniensis* in the shape of the proboscis and the collar and their relative sizes, in the clear-cut demarcation of the collar from the trunk, in the shape and appearance of the branchiogenital region and in the less-developed genital ridges, which are like alae in *G. sarniensis*. In internal anatomy, *G. barnharti* differs from *G. sarniensis* in the following: the circular-muscle layer of the proboscis is much thicker in *G. barnharti*; in the possession of a small dorsal glomerulus, much narrower stomochordal lumen and flattened forepart of stomochord; middorsal proboscis pore; extension of the skeletal keel into the collar region, absence of ventral septum in collar, absence of continuous canal in the collar nerve cord, many more dorsal roots, absence of posterior neuropore, and in the position of the gonads.

The above facts show that the present species does not resemble sufficiently well either *G. minutus* or *G. sarniensis* for it to be included under those species. Still less does it resemble any of the other species of the genus. The defining characters of *Glossobalanus barnharti* are:

- flattened forepart of stomochord
- a single midventral blind pouch to the stomochord
- peduncle skeletal keel is deep and narrow, extending well into collar
- collar nerve cord is flattened
- eight or nine dorsal roots in collar

Discussion

On the west coast of North America, the genus *Glossobalanus* has been taken once in the Salish Sea at Nanaimo, British Columbia (Willey 1931), once in Coos Bay, Oregon and three times in La Jolla, California, representing four different species. It is likely to turn up anywhere. Probably the 'ptychoderids' reported by Woodwick (1955), in three dredge samples at 40–90 m depth from San Pedro Basin, belong to this genus. The habitats include both sand flats and rocky rubble on the outer coast. For a detailed discussion of the distribution of North American acorn worms, including the new species described here, see Cameron *et al*. (2010).

Dichotomous keys to the genera of the enteropneust families Harrimaniidae (Deland *et al*. 2010) and Spengelidae (Cameron and Perez, 2012) have been developed. The deep-sea family Torquaratoridae consists of five well-supported clades (Osborn *et al*. 2012), and the corresponding morphological descriptions are presently being assembled in the laboratory of professor Nicholas Holland (Scripps Institute of Oceanography). Here we provide a taxonomic key to the family Ptychoderidae.

A key to the genera of the family Ptychoderidae.

Stomochord with no vermiform process; pericardium simple; ventral part of pharynx large and sometimes more or less separated from the branchial part; liver sacs, synapticles, and nerve roots usually present. **Ptychoderidae**

(i)   Genital wings well developed
       (a) Atrium opening by long slits . . . . *Ptychodera*
       (b) Atrium opening by small pores   . . . . *Balanoglossus*
(ii)   Genital wings hardly developed . . . . *Glossobalanus*

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